PROCEEDINGS OF

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Section B.—Biological Sciences.

On the Occurrence of Multinucleate Cells in Vegetative Tissues.

By Rudolf Beer, B.Sc., F.L.S., and Agnes Arber, D.Sc., F.L.S., Fellow of Newnham College, Cambridge.

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(Plate 1.)

Historical Introduction.

The typical animal or vegetable cell is now universally held to consist of a protoplasmic body, either naked or enclosed in a cell membrane and containing, as its most essential constituent, a single nucleus. This view as to the uninucleate character of the vegetable cell was first definitely formulated by Nägeli in 1844. A few well-defined exceptions to this rule, such as pollen grains, embryo sacs, etc., were recognised by Nägeli himself, and during the three-quarters of a century which has elapsed since the publication of his work, several other instances of multinucleate cells have been observed by Schmitz, Treub, Johow, Strasburger, Grant, and others. These cases, in which a plurality of nuclei was seen to occur in the cell, were however regarded as isolated exceptions to an otherwise universal rule, and of no general significance.

In 1914 Dr. McLean* observed cells with more that one nucleus in the tissues of certain water plants, while in 1915 Miss Prankerd† published an account of her researches on multinucleate cells. She recorded the occurrence of multinucleate cells in 36 species of plants "widely separated in

* McLean, R. C., "Amitosis in the Parenchyma of Water-Plants," 'Proc. Camb. Phil. Soc.,' vol. 17, pp. 380–382 (1914), 1 text-fig.

† Prankerd, T. L., "Notes on the Occurrence of Multinucleate Cells," 'Ann. Bot.,' vol. 29, pp. 599–604 (1915), 8 text-figs.

habit, habitat, and systematic position," including both Vascular Cryptogams and Angiosperms. Miss Prankerd considers, like nearly all of those who have written upon this subject, that the plurality of nuclei arises by amitosis. With regard to the ultimate fate of the nuclei, she does not believe it probable either that all the nuclei but one degenerate in each cell, or that they fuse with one another. She is inclined to suppose that walls may ultimately be formed between the daughter nuclei which have arisen by amitosis.

In the same issue of the 'Annals of Botany' as that in which Miss Prankerd's account appeared, we published a preliminary statement of our results upon the same subject.* It is unnecessary to refer to this at length, but we may recall that we recorded a plurality of nuclei in the young parenchymatous tissues of 76 species, chiefly Angiosperms, but including also a Gymnosperm and a Vascular Cryptogam. This phenomenon seemed to us so widespread that we suggested the possibility that a binucleate or multinucleate stage might often intervene as a normal phase of development between the meristematic and adult conditions. The main difference between our results and those of previous writers is that, according to our observations, the plurality of nuclei normally arises by karyokinesis and not by amitosis. Moreover, there are certain peculiarities in the phenomena associated with this mitosis to which we called attention in our note, and which we propose to describe more fully in the present paper.

The Occurrence of the Multinucleate Phase.

Binucleate or multinucleate cells have been observed by us in the vegetative tissues of 177 species representing 60 families. They have been found in each of the five classes of living Pteridophyta (*e.g.*, *Equisetum*, Plate 1, fig. 25, and *Lygodium*, Plate 1, fig. 28), and are of very general occurrence in the Gymnosperms (*e.g.*, *Larix*, Plate 1, fig. 23, and *Cryptomeria*, Plate 1, fig. 27), Monocotyledons, and Dicotyledons. The cases in which we have observed them are enumerated in the following list. An asterisk before the name of a species denotes that it is a case in which "phragmospheres" have been observed (see p. 10). All the observations were made on our own preparations, except in the case of those species to which the initials (E.S.) are appended; these have been examined in the late Miss Ethel Sargant's collection of seedling slides.

* Beer, R., and Arber, A., "On the Occurrence of Binucleate and Multinucleate Cells in Growing Tissues," 'Ann. Bot.,' vol. 29, pp. 597–8 (1915).

List of Species in which Binucleate or Multinucleate Cells have been observed in Vegetative Tissues.

Family.	Species.	Region in which cells containing more than one nucleus occur.
PTERIDOPHYTA.		
Filicales.		
Polypodiaceæ	*Aspidium filix mas*, L. var. *capitatum*	Prothallus.
	Athyrium filix femina, L. Roth var. *grandiceps*	Prothallus.
	Scolopendrium vulgare, Sm. var. *ramosissimum*	Prothallus.
Schizæaceæ	**Lygodium japonicum*, Sw.	Mesophyll of climbing frond.
Equisetales.		
Equisetaceæ	**Equisetum limosum*, L.	Cortex and pith of fertile stem.
	**E. maximum*, Lmk.	Cortex and pith of fertile stem.
Lycopodiales.		
Selaginellaceæ ...	**Selaginella Wildenovii*, Baker...	Cortex of stem.
Psilotales.		
Psilotaceæ	**Psilotum triquetrum*, Sw.	Cortex of stem.
Isoëtales.		
Isoëtaceæ	*Isoëtes, sp.	Lacunar cortex of stem.
GYMNOSPERMEÆ.		
Coniferæ.		
Taxaceæ	**Taxus baccata*, L.	Pith and leaf bases (including epidermis) of shoot.
Pinaceæ.		
Araucarieæ......	*Araucaria Bidwillii*, Hook.	Leaf bases of shoot.
	**A. excelsa*, R. Br..................	Pith of shoot.
Abieteæ	*Larix europæa*, DC.	Leaf bases of shoot.
	Pinus silvestris, L.	Leaf bases and cortex of seedling stem.
Taxodieæ	**Cryptomeria japonica*, (L.) Don.	Leaf bases of shoot.
ANGIOSPERMEÆ.		
Monocotyledoneæ.		
Pandanaceæ	**Pandanus Veitchi*, Hort.	Cortex and stelar tissue of root.
Sparganiaceæ......	*Sparganium ramosum*, Curt.	Ground tissue of inflorescence axis.
Aponogetonaceæ	*Aponogeton distachyum*, Thunb.†	Lacunar cortex of inflorescence axis; mesophyll of lamina and ground tissue of petiole; mesophyll of spathe.
Alismataceæ	**Alisma Plantago*, L.	Ground tissue of petiole and inflorescence axis.
Hydrocharitaceæ	**Elodea canadensis*, Michx.‡	Cortex and epidermis of shoot.
	**Stratiotes aloides*, L.	Cortex of root; cortex of stolon; mesophyll of leaf.
Gramineæ	*Arrhenatherum avenaceum*, Beauv.	Leaf sheath and ground tissue of stem.
	**Arundinaria* "*palmata*"	Ground tissue of stem.
	**Avena sativa*, L.	Leaf sheath and ground tissue of stem.
	Bambusa metake, Sieb. (= *Arundinaria japonica*, Sieb. et Zucc.)	Ground tissue of stem.
	**Bambusa* "*anceps*"	Ground tissue of vegetative stem and cortex of root.

† McLean, R. C. (*l.c.*) records the occurrence of binucleate cells in the cortex of *Aponogeton*, sp.
‡ This case was recorded by McLean, R. C. (*l.c.*), but phragmospheres were not observed.

List of Species—(*contd.*)

Family.	Species.	Region in which cells containing more than one nucleus occur.
ANGIOSPERMEÆ. *Monocotyledoneæ.*		
Gramineæ	*Coix Lacryma-Jobi*, L.	Ground tissue of stem.
	Dactylis glomerata, L.	Leaf sheath.
	Festuca, sp.	Ground tissue of stem and leaf sheath.
	Hordeum vulgare, L.	Leaf sheath.
	Leersia oryzoides, Sw.	Mesophyll of first plumular leaf and cortex of mesocotyl (E.S.).
	Lolium italicum, A. Br.	Mesophyll of first plumular leaf (E.S.).
	**Miscanthus sacchariflorus*, Hack.	Ground tissue of stem.
	Secale cereale, L.	Leaf sheath.
	Sorghum vulgare, Pers.	Cortex of mesocotyl, and mesophyll of coleoptile and first plumular leaf (E.S.).
	Triticum vulgare, Vill.	Leaf sheath.
	**Zea Mays*, L.	Cortex of mesocotyl and mesophyll coleoptile (E.S.); and leaf sheath and ground tissue of stem; plerome, periblem and piliferous layer of root.
Cyperaceæ	**Cyperus Papyrus*, L.	Ground tissue of young axis and leaf bases.
Araceæ	**Anthurium Martianum*, C. Koch et Kolb	Cortex of aerial root.
	**A. scandens*, Engler (= *A. violaceum*, Schott)	Ground tissue of stem and leaf sheath. Epidermis of leaf sheath.
	**Monstera deliciosa*, Liebm.	Cortex of aerial root.
	**Philodendron latifolium*, C. Koch	Cortex of aerial root.
Commelinaceæ	**Tradescantia virginiana*, L.	Ground tissue of stem.
	**T. fluminensis*, Vell.	Ground tissue of stem.
Liliaceæ	**Allium angulosum*, L.	Cotyledon (E.S.).
	**A. ascalonicum*, L.	Cotyledon (E.S.).
	**A. Cepa*, L.	Cotyledon (E.S.).
	**A. neapolitanum*, Cyr.	Cotyledon (E.S.).
	**A. Porrum*, L.	Cotyledon (E.S.).
	**Aloe striata*, Haw. (*Hanburyana*)	Mesophyll of first plumular leaf (E.S.).
	**A. sp.* (near *Caspari*)	Mesophyll of first plumular leaf (E.S.).
	Anthropodium cirrhatum, R. Br.	Mesophyll of first plumular leaf (E.S.).
	**Asparagus officinalis*, L.	Cortex, ground tissue and pith of vegetative axis; also (rarely) epidermis and xylem parenchyma.
	**Asphodeline liburnica*, Reichb.	Cotyledon, hypocotyl and mesophyll of first leaf (E.S.).
	Asphodelus albus, Willd.	Cotyledon and first plumular leaf (E.S.).
	A. fistulosus, L.	Hypocotyl (E.S.).
	Bloomeria aurea, Kellogg	First plumular leaf (E.S.).
	Brodiæa lactea, S. Wats.	Mesophyll of first plumular leaf (E.S.).
	Bulbine annua, Willd.	Hypocotyl and mesophyll of first plumular leaf (E.S.).
	Chlorogalum pomeridianum, Kunth.	Mesophyll of first plumular leaf (E.S.).
	Colchicum autumnale, L.	Mesophyll of first plumular leaf (E.S.).
	Convallaria majalis, L.	Pith and cortex of inflorescence axis.
	Cordyline australis, Hook. f.	Mesophyll of first plumular leaf (E.S.).
	**Dipcadi serotinum*, Medic.	Cotyledon (E.S.).

List of Species—(*contd.*)

Family.	Species.	Region in which cells containing more than one nucleus occur.
ANGIOSPERMEÆ.		
Monocotyledoneæ.		
Liliaceæ	**Dracæna Draco*, L.	Cotyledon and mesophyll of first plumular leaf (E.S.) and pith of root of mature plant.
	**Eremurus himalaicus*, Baker	Pith, ground tissue and cortex of inflorescence axis, and mesophyll of leaf.
	**E. turkestanicus*, Regel	Mesophyll of first plumular leaf (E.S.).
	Eucomis nana, L'Hérit.	Mesophyll of first plumular leaf (E.S.).
	Galtonia candicans, Decne.	Cotyledon, hypocotyl and mesophyll of first plumular leaf (E.S.).
	**Gasteria disticha*, Haw.	Mesophyll of first plumular leaf (E.S.).
	**Hemerocallis fulva*, L.	Ground tissue of inflorescence axis and mesophyll of leaf.
	Hyacinthus romanus, L.	Cotyledon and cotyledonary node (E.S.)
	**Kniphofia* (garden var.)	Ground tissue of inflorescence axis.
	Muscari comosum, Mill.	Cotyledon (E.S.).
	M. neglectum, Guss.	Cotyledon (E.S.).
	M. racemosum, Mill.	Pith, cortex and starch sheath of inflorescence axis.
	**Nothoscordum fragrans*, Kunth.	Ground tissue of inflorescence axis.
	Ornithogalum exscapum, Tenore	Cotyledon (E.S.).
	**Polygonatum multiflorum*, All.	Ground tissue of inflorescence axis.
	Scilla festalis, Salisb.	Mesophyll of plumular leaf (E.S.).
	**S. hispanica*, Miller	Pith and cortex of inflorescence axis and mesophyll of leaf.
	S. peruviana, L.	Cotyledon, hypocotyl and first plumular leaf (E.S.).
	S. sibirica, Andr.	Cotyledon (E.S.).
	Tulipa (garden var.)	Ground tissue of floral axis.
	Veltheimia viridifolia, Jacq.	Cotyledon and mesophyll of first plumular leaf (E.S.).
	**Yucca aloifolia*, L.	Mesophyll of first plumular leaf (E.S.).
	**Y. arborescens*, Trelease	Mesophyll of first plumular leaf and hypocotyl (E.S.)
	Y. gloriosa, L.	Cotyledon, primary root and mesophyll of first plumular leaf (E.S.).
Amaryllidaceæ	**Agave spicata*, Cav.	Cotyledon, hypocotyl and plumular leaves (E.S.).
	**Bravoa geminiflora*, Lex.	Mesophyll of first plumular leaf (E.S.).
	Doryanthes Palmeri, W. Hill.	Hypocotyl, and mesophyll of first plumular leaf (E.S.).
Dioscoreaceæ	**Tamus communis*, L.	Pith and cortex of axis.
Iridaceæ	*Crocus vernus*, All. (garden var.)	Parenchyma of leaves and perianth of bud.
	Iris Boissieri, Henriq.	Mesophyll of first plumular leaf (E.S.).
	**I. sibirica*, L.	Mesophyll of plumular leaf (E.S.).
	**I. sp.*	Mesophyll of plumular leaf (E.S.).
Zingiberaceæ	**Hedychium*, sp.	Ground tissue of axis, mesophyll of leaf sheath, and cortex of root.
Orchidaceæ	*Calanthe Veitchii* (garden form)	Leaf bases.
	Neobenthamia gracilis, Rolfe	Root cortex.
Dicotyledoneæ		
Betulaceæ	*Corylus Avellana*, L.	Pith and cortex of stem.
Moraceæ	*Morus nigra*, L.†	Pith and cortex of axis.

† This case has already been recorded by Prankerd, T. L. (*l.c.*), but phragmospheres were not noted.

List of Species—(*contd.*)

Family.	Species.	Region in which cells containing more than one nucleus occur.
ANGIOSPERMEÆ.		
Monocotyledoneæ.		
Polygonaceæ	**Polygonum cuspidatum*, Sieb. et Zucc.†	Pith, cortex and epidermis of the stem.
	**P. senegalense*, Meissn.	Pith of stem.
	**P. Weyrichii*, F. Schmidt.........	Pith of inflorescence axis.
	Rheum "*corallinum*"	Pith of inflorescence axis.
	R. Rhaponticum, L.	Petiole.
	**Rumex obtusifolius*, L.	Pith and cortex of stem.
Nymphæaceæ	*Cabomba*, sp..........................	Lacunate cortex of stem and mesophyll of leaf.
	**Victoria regia*, Lindl.	First internode of seedling, petiole of leaf, peduncle and root cortex.
Ranunculaceæ ...	**Aconitum Napellus*, L.	Cortex and pith of inflorescence axis and vegetative axis.
	**Delphinium* (garden var.).........	Pith and cortex of stem.
	**Thalictrum flavum*, L.	Pith of inflorescence axis.
Papaveraceæ	**Chelidonium majus*, L.	Pith of stem.
	**Glaucium flavum*, Crantz	Pith of inflorescence axis.
Cruciferæ	**Brassica oleracea*, L. ("Savoy")	Pith of elongating stem of "bolting" plant.
	**Lepidium Draba*, L.	Pith and cortex of stem.
	**Sisymbrium Alliaria*, Scop.	Pith and cortex of inflorescence axis.
	**S. officinale*, Scop.	Pith of stem.
Resedaceæ	**Reseda lutea*, L.	Pith of inflorescence axis.
Saxifragaceæ	**Philadelphus coronarius*, L.......	Pith of stem.
	**Ribes rubrum*, L.	Pith and cortex of vegetative axis.
Rosaceæ	**Kerria japonica*, DC.	Pith of inflorescence axis.
	**Pyrus Malus*, L.	Pith and cortex of stem.
	**Rosa* (garden var.)..................	Pith and cortex of stem.
	**Rubus fruticosus*, L.	Pith of stem.
Leguminosæ	**Galega officinalis*, L.	Cortex and pith of stem.
	**Phaseolus multiflorus*, Willd. ...	Pith of stem.
Geraniaceæ.........	**Pelargonium zonale*, L'Hérit. ...	Pith and cortex of inflorescence axis.
	**Tropæolum majus*, L.	Stem, cotyledonary node, petiole of cotyledon, hypocotyl, cortex and pith of root.
Buxaceæ	**Buxus sempervirens*, L............	Cortex and pith of vegetative axis.
Anacardiaceæ......	**Rhus Coriaria*, L.	Pith of inflorescence axis.
Aceraceæ	**Acer campestre*, L.	Pith of vegetative axis.
Hippocastanaceæ	**Æsculus Hippocastanum*, L.‡ ...	Pith and cortex of stem and inflorescence axis.
Vitaceæ	**Vitis inconstans*, Miq. (= *Ampelopsis Veitchii*)	Cortex and pith of vegetative axis.
	V. pterophora, Baker	Cortex and pith of aerial root.
Malvaceæ	**Althæa rosea*, Cav.	Cortex and pith of axis.
Oenotheraceæ......	**Œnothera longiflora*, L.	Cortex and pith of stem.
Hippuridaceæ......	**Hippuris vulgaris*, L.§	Cortex of stem.
Araliaceæ	**Hedera Helix*, L.‖	Pith and cortex of stem.

† This appears to be the case recorded under the name of *P. Sieboldii*, by Grant, but phragmospheres were not observed by him. See Grant, A. E., "The Multinucleated Condition of the Vegetable Cell," 'Trans. Bot. Soc. Edinb.,' vol. 16, pp. 38–52 (1886), two Plates.

‡ This case was recorded by Prankerd, T. L. (*l.c.*), but phragmospheres were not observed.

§ This case was recorded by McLean, R. C. (*l.c.*), but phragmospheres were not observed.

‖ Binucleate cells were recorded in the petiole of this plant by Prankerd, T. L. (*l.c.*) but phragmospheres were not observed.

List of Species—(*contd.*)

Family.	Species.	Region in which cells containing more than one nucleus occur.
ANGIOSPERMEÆ. *Monocotyledoneæ.*		
Umbelliferæ	**Ægopodium Podagraria*, L.	Pith and cortex of inflorescence axis.
	**Anthriscus sylvestris*, Hoffm.	Pith and cortex of inflorescence axis.
	**Carum Petroselinum*, Benth. et Hook.	Pith and cortex of inflorescence axis.
	**Caucalis daucoides*, L.	Cortex and pith of axis.
	**Heracleum giganteum*, Fisch.	Pith and cortex of inflorescence axis.
	**Œnanthe fistulosa*, L.	Pith of stem and cortex of root.
	**Siler trilobum*, Crantz	Petiole; pith and cortex of inflorescence axis and of stalks of partial umbels.
	**Smyrnium Olusatrum*, L.	Pith and cortex of inflorescence axis.
Ericaceæ	**Arbutus Unedo*, L.	Pith and cortex of stem.
Oleaceæ	**Fraxinus excelsior*, L.†	Pith and cortex of vegetative axis.
	**Syringa vulgaris*, L.†	Pith and cortex of vegetative axis.
Convolvulaceæ	**Calystegia silvatica*, Choisy	Pith and cortex of vegetative axis.
Polemoniaceæ	*Phlox* (garden var.)	Pith of vegetative axis.
Labiatæ	**Stachys sylvatica*, L.	Pith and cortex of stem.
Solanaceæ	**Solanum nigrum*, L.	Pith of stem.
Scrophulariaceæ	**Antirrhinum* (garden var.)	Pith and cortex of vegetative axis.
	**Calceolaria Pavonii*, Benth.	Pith and cortex of stem.
	**Digitalis purpurea*, L.	Pith and cortex of inflorescence axis.
Acanthaceæ	**Acanthus mollis*, L.	Parenchyma of petiole and inflorescence axis.
Plantaginaceæ	**Plantago major*, L.	Pith of inflorescence axis.
Caprifoliaceæ	**Sambucus nigra*, L.	Epidermis, cortex and pith of stem.
	**Viburnum Lantana*, L.	Pith of stem.
	**Symphoricarpos*, sp.	Pith of stem.
Dipsacaceæ	**Dipsacus laciniatus*, L.	Pith of vegetative axis.
Cucurbitaceæ	**Bryonia alba*, L.	Ground tissue of vegetative axis.
	**B. dioica*, Jacq.	Ground tissue between bundles of vegetative axis.
	**Thladiantha dubia*, Naud.	Cortex and ground tissue of vegetative axis.
	**T. Oliveri*, Cogn.	Pith and cortex of vegetative axis.
Compositæ	**Centaurea nigra*, L.	Pith of stem.
	**Chrysanthemum Parthenium*, Bernh.	Cortex and pith of vegetative axis.
	**Crepis taraxacifolia*, Thuill.	Pith of stem.
	**Dahlia* (garden var.)	Cortex and pith of stem.
	**Doronicum* sp. (probably *Pardalianches*, L.)	Cortex and pith of inflorescence axis.
	**Helianthus Nuttallii*, Torr. et Gray	Cortex and pith of inflorescence axis.
	**H. tuberosus*, L.	Cortex and pith of vegetative axis.
	**Solidago stricta*, Ait.	Pith of vegetative axis.
	**Taraxacum officinale*, Weber	Cortex and pith of inflorescence axis.
	**Tragopogon pratensis*, L.	Cortex and pith of inflorescence axis.

† The occurrence of binucleate cells in these species was recorded by Prankerd, T. L. (*l.c.*), but phragmospheres were not observed.

It will be seen from the above list that the multinucleate condition has been found in the most various vegetative organs. It is most frequently and

characteristically met with in the stem, but it has also been observed in many roots—subterranean, aerial, and aquatic (*e.g.*, *Stratiotes*, Plate 1, fig. 29). It occurs in many leaf structures, such as the sheathing leaf bases of a number of grasses (*e.g.*, *Zea*, Plate 1, fig. 3, and *Avena*, Plate 1, fig. 18) and in the basal zone of the perianth of *Crocus*. We have frequently met with binucleate cells in the cotyledon, plumular leaf, mesocotyl and hypocotyl of seedlings. The range of tissues concerned is also very wide, including pith, cortex, epidermis and stele of stem, mesophyll of leaf, cortex and stele of root.

In all the cases in which we have observed multinucleate cells, they are characteristic of young tissues which are actively carrying on the processes of life. For example, in the stem the multinucleate cells first appear in a region just behind the actual meristematic tissue, where preparation is being made for the growth in length of the organ. In *Helianthus Nuttallii* and *Syringa vulgaris* binucleate cells make their first appearance 0·1 mm. behind the stem apex.

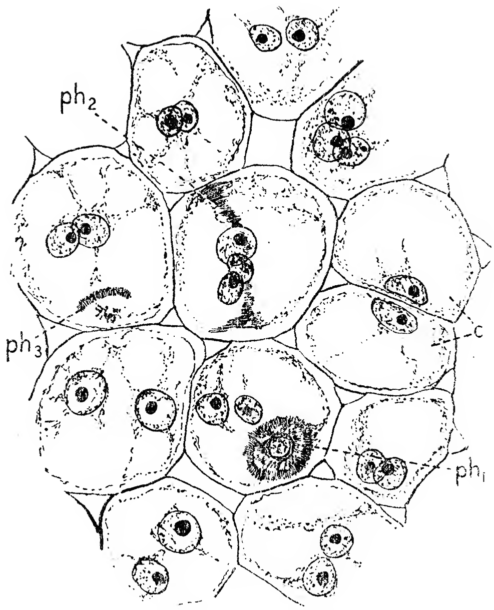
In those stems which have active tissue, capable of carrying out processes of growth, situated at the nodes (such as the stems of Gramineæ and of *Tradescantia*) the multinucleate cells occur predominantly at these spots.

The existence of the multinucleate phase is very easily demonstrated. If for instance, sections be made across the "heads" of *Asparagus*, at the stage at which they are cut for market, preparations such as those shown in text-figs. 1 and 2 will be invariably obtained.

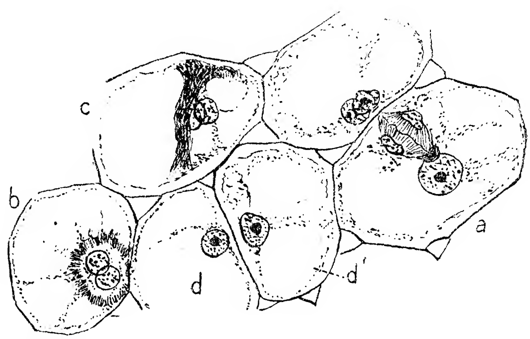
The number of nuclei present in a multinucleate cell varies greatly in different species. Most frequently the cells are binucleate, but in many cases three, four, or even more nuclei may occur in a cell. In the tissues just above a node in the stem of *Zea Mays* (Plate 1, fig. 4) as many as 12 nuclei may sometimes be counted in a single cell, whilst in the young inflorescence axis of *Anthriscus sylvestris*, cells containing 8, 9, and 10 nuclei are not uncommon (*e.g.*, Plate 1, fig. 13).

The Origin of the Multinucleate Phase.

Nearly all previous writers have been agreed in holding the view that the origin of the multinucleate condition in a cell is due to the amitotic division of the originally single nucleus. Johow, Strasburger, Grant, McLean and Prankerd have all expressed this opinion; Smolák and Němec are almost alone in having observed a definite instance in which plurality of nuclei became established in the cell by the karyokinetic division of the primary nucleus. They found this to be the case in the plerome cells



TEXT-FIG. 1.—*Asparagus officinalis*, L. Cells of ground tissue between the bundles from a transverse section across the "head" of a shoot gathered on May 6, 1916. (×535 *circa*.) One phragmosphere (ph_1) is seen at an early stage of development in a cell containing two resting nuclei; a second (ph_2) is at a more advanced stage of development in a cell containing one resting nucleus; a third (ph_3) is only seen in fragmentary tangential section. Two cells (c) are uninucleate; these are obviously young cells which have recently divided from one another.



TEXT-FIG. 2.—*Asparagus officinalis*, L. Cells of ground tissue between the bundles from a transverse section 3–4 mm. below the base of the "head" of the same shoot as that from which the section represented in Text-fig. 1 was taken. (×535 *circa*.) Progressive wall formation is going on in cell *a*, simultaneously with phragmosphere formation in cells *b* and *c*. Just as in Text-fig. 1, the only uninucleate cells are a pair which have been recently formed (*d* and *d'*).

and vessel rudiments of the root of several species of *Euphorbia* and *Ricinus*, but these plants were regarded as exceptions to the general rule according to which multinucleate cells arise by amitotic divisions of the cell nucleus.

We have gone very carefully into the question of the origin of the plurality of nuclei in the multinucleate cells and in more than 100 species we have been able to satisfy ourselves beyond doubt that the several nuclei have arisen by mitotic division. No single instance of direct nuclear division has ever been observed by us in these young active tissues.

The Formation of Phragmospheres.

Certain peculiarities are found in the course of these mitotic divisions which appear to be characteristic of multinucleate cells. We briefly referred to these peculiarities in our preliminary note, when we described them as giving rise to an appearance of a cell lying within another cell. We have now observed this modified form of mitosis in the young vegetative tissues of some 120 species belonging to more than 50 families, including representatives of Vascular Cryptogams, Gymnosperms, and Angiosperms. We therefore feel that we are justified in regarding the process observed as being possibly almost universal in these young tissues.

In cells which are about to become binucleate, the mitosis proceeds normally up to the spindle stage and the cell plate makes its appearance as usual; it does not, however, give rise to any cell membrane. It is apparently resorbed, and the whole phragmoplast* with the associated cytoplasm becomes transformed into a hollow sphere which encloses the daughter nuclei; this sphere gradually increases in diameter, ultimately becoming co-extensive with the cytoplasm lining the cell wall. For this hollow shell, which represents the later development of the phragmoplast, and which, in the case of binucleate cells, replaces the normal apparatus of wall formation, we propose the term "phragmosphere."† The spherical shape is not fully exhibited unless the karyokinetic figure occurs in the centre of the cell. If, as often happens, it occupies a position close to a wall, the sphere at an early stage of its expansion fuses on one side with the primordial utricle and thus loses its symmetry. As the phragmosphere extends, the paired nuclei gradually move apart from one another, while they develop in normal fashion and ultimately assume the ordinary characters of resting nuclei.

The young inflorescence axis of *Anthriscus sylvestris* has furnished some of

* See Errera, L., "Ueber Zellformen und Seifenblasen" (Versamml. Deutsch. Naturfor. u. Aerzte in Wiesbaden), 'Bot. Centralbl.,' vol. 34, pp. 395–398 (1888).

† *Φραγμός* = hedge, fence, etc.; *σφαῖρα* = sphere.

the most characteristic examples of phragmospheres for critical examination (see Plate 1, figs. 7–12). In many cases only a single phragmosphere occurs in a cell in association with a single dividing nucleus; in others two phragmospheres with both pairs of nuclei at the same stage of division may be observed in a single cell, or one phragmosphere with its nuclei in telophase, a second nucleus in the anaphase of division, and a third nucleus in the resting condition may all occur together in one cell. The history of the phragmosphere in *Anthriscus* is precisely as outlined in the preceding paragraph. By the disintegration of the spindle and the disappearance of the cell plate before it has split into two, the two daughter nuclei, in telophase, are left surrounded by a mass of cytoplasm which is partly kinoplasmic in origin. After a time this cytoplasmic aggregate becomes slightly vacuolar in its interior; the vacuole gradually enlarges, pushing a hollow shell of dense cytoplasm more and more nearly towards the outer boundary of the cell until it merges into the peripheral protoplasm. The spindle fibres break down very completely in *Anthriscus* and the phragmospheres show very little of that appearance of being composed of radiating fibres which is more clearly seen in some of the other plants which we have examined (*e.g.*, *Zea Mays*, Plate 1, figs. 1 and 2). In *Anthriscus* the phragmosphere has a densely granular constitution.

No previous writer appears to have recognised the existence of phragmospheres, or of anything like them, in ordinary vegetative tissues, but more than one observer has recorded the appearance, within the endosperm, of structures which may possibly be either related to phragmospheres or identical with them. Němec,* for instance, describes in the endosperm of *Secale cereale*, certain bodies, which he calls "Kerntaschen," which bear some resemblance to phragmospheres, but he did not follow out their history in detail. Again, certain figures of endosperm cells, taken by the observers to suggest end views of nuclear spindles, may possibly indicate phragmospheres which have been drawn without a full understanding of their nature.† Among Gymnosperms, Lawson's‡ account of the formation of binucleate cells in the endosperm of *Cryptomeria japonica* suggests the occurrence of structures somewhat resembling phragmospheres. Beyond these few notes, whose relevance is doubtful, we have found nothing in the literature which could possibly be construed as a reference to phragmospheres.

* Němec, B., 'Das Problem der Befruchtungsvorgänge,' Berlin, pp. 112, 113 (1910).

† Němec, B., *ibid.*, Plate 2, fig. 50; Buscalioni, L., 'Annuario del R. Istituto Bot. di Roma,' vol. 7 (1898), Pl. 17, fig. 77B and Pl. 20, fig. 140.

‡ Lawson, A. A., 'Ann. Bot.,' vol. 18, p. 427 (1904), Plate 29, figs. 28–32.

The Fate of the Nuclei.

The fate of the nuclei in multinucleate cells varies in the different species.

In some cases, the plurality of nuclei may persist to a late stage. Thus in the stem of *Syringa vulgaris* binucleate cells still occur in the perimedullary zone as far as 93 cm. from the apex, whilst in *Rosa* binucleate cells were still seen in the cortex of a two-year old stem (Plate 1, fig. 17).

In other cases one or more of the nuclei may undergo degeneration (*e.g.*, *Zea Mays*, Plate 1, fig. 6). This can be well seen in the flowering axis of *Hemerocallis fulva.* In material gathered on May 21 and 24 (1916) the region of the axis just below the base of the young inflorescence showed numerous cases of cells in which one nucleus was normal, while the second was obviously degenerating. The degenerating nuclei are contracted and irregular in form ; they seem to be structureless, since they stain more or less homogeneously, while the normal nuclei accompanying them are granular in appearance with a number of nucleoli. Further down the axis the cells are mostly uninucleate.

Although we have spent much time on the matter, no instances of nuclear fusions, as factors in the reduction of the number of the nuclei, have been observed. Neither has any evidence been forthcoming of the existence of a belated cell-division leading to the separation of the nuclei of a multinucleate cell by the interpolation of new cell walls between them. Lobed nuclei have been observed in several instances, *e.g.*, *Helminthostachys*, Plate 1, fig. 24, but a careful and critical examination of these cases has convinced us that these have neither the significance of nuclear fusions nor of amitotic divisions. They merely represent a change of form which is the expression of metabolic activities taking place in the cell. *Statiotes aloides*,* *Asparagus officinalis*, and *Tradescantia virginiana* furnish instances of such lobed nuclei.

The case of *Tradescantia* is particularly noteworthy, since it has long been regarded as one of the classical instances of nuclear amitosis in the higher plants. We have, however, found unmistakable evidence that the plurality of nuclei in the stems of this plant arises by mitotic division accompanied by the occurrence of characteristic phragmospheres. The lobing of the nuclei is here, as elsewhere, a mere change of form, which rarely if ever bears any relation to their multiplication.

After our work on *Tradescantia* was completed, a recent paper by

* In a former paper by one of us (Arber, A., 'Proc. Camb. Phil. Soc.,' vol. 17, p. 369 (1914)), the origin of the binucleate phase in the roots of *Stratiotes* was wrongly attributed to amitosis. Further work has shown that the binucleate condition arises by karyokinesis accompanied by phragmosphere formation (Plate 1, fig. 29), and that the lobing is a distinct phenomenon.

Schürhoff came to our notice. This will necessitate a somewhat fuller treatment of this plant than we anticipated, or than could be incorporated in the present account. We propose, therefore, to give here only the foregoing brief reference to our results and to reserve the description of further details, together with a discussion of Schürhoff's work, for a separate communication.

The Significance of the Multinucleate Phase.

In the foregoing pages we have called attention to a long list of species in which multinucleate cells occur, not as a chance phenomenon, but as a normal and definite stage in the development of their parenchymatous tissues. We may add that not a single species of flowering plant has been met with in our work in which, after a careful examination, multinucleate cells have not been found to occur to a greater or less extent. We therefore feel that we are justified in concluding that the occurrence of a multinucleate phase is a normal feature in the growth of the majority of the higher plants. It is a phase which in general succeeds the meristematic activity of the cells, and precedes their period of maximum growth. The phase may endure throughout the whole period of growth, and, indeed, traces of it may be found in quite old tissues, or it may give place to a uninucleate condition at an extremely early stage. From the point of view of the development of the individual cell, the multinucleate phase may be regarded as a stage at which the cytoplasm has either temporarily or permanently lost its ability to divide, whilst this power is still retained by the nucleus. A number of cases are already known, both in zoological and botanical literature, in which the cytoplasm and nucleus of a cell are unequally affected by various agencies which exert an influence upon the course of a mitotic division. Němec's* well-known work dealing with the effect of chloral hydrate and other such substances upon cell division is a familiar example, whilst Demoor's† earlier observations upon the effect of chloroform upon the hairs of *Tradescantia* showed that the streaming movements of the cytoplasm are arrested before the mitotic division of the nucleus is interfered with. Loeb‡ has recorded the interesting fact that, at a certain concentration of sea-water (either with sodium chloride, or preferably with magnesium chloride), the cytoplasmic divisions are arrested in a freshly fertilised egg of a Sea Urchin, while the nuclear divisions continue.

* Němec, B. 'Das Problem der Befruchtungsvorgänge,' Berlin, 1910.

† Demoor, J., 'Archives de Biologie,' vol. 13, pp. 163–244 (1895).

‡ Loeb, J., 'Archiv f. Entwickelungsmechanik,' vol. 2, pp. 298–300 (1896).

Raciborski* found that, when an old culture of *Basidiobolus ranarum* was transferred to a 10 per cent. solution of glycerine and the temperature raised to 30° C., cell-division was inhibited, whilst nuclear division continued.

All these observations refer to the establishment of the multinucleate condition as a pathological phenomenon due to the action of abnormal conditions upon the cell. In the series of cases dealt with in the present paper, the phenomenon is entirely due to normal agencies arising in the ordinary life of the plant. The transition from the meristematic to the adult condition apparently affects the cytoplasm first and the nucleus only at a later period. It is interesting to note, however, that the cytoplasm is still capable of taking a considerable share in the mitosis, since a perfectly normal spindle is differentiated which effects the regular distribution of the daughter chromosomes to the poles. A cell plate even is formed at the equator of the spindle, but, so far as could be seen, this plate never splits, and it is apparently at this point that the cytoplasmic mechanism breaks down.

This independent susceptibility of cytoplasm and nucleus is probably not without its significance in the life of the plant.

As we have already stated, the multinucleate phase reaches its most characteristic expression just previous to the maximum period of growth undergone by the region of the stem, leaf, or root in which it occurs. At such a time metabolic activities must be running high in these cells and it is very probable that the multinucleate condition is in some way directly associated with the elaboration of material required for their growth.

Elsewhere we have abundant evidence that the nucleus plays an important *rôle* in cell metabolism, and a large number of facts have been recorded which tend to show that an active interchange of influences, probably both material and dynamic, takes place between the nucleus and the cytoplasm during the elaboration of materials by the cell. This interchange is undoubtedly facilitated by an increase in the nuclear surface.

A number of cases are known in the animal organism in which cells engaged in intense secretory activity exhibit a greatly enlarged nuclear surface. The silk-glands of various insect larvæ, examined by Meckel,† Zaddach,‡ and Korschelt,§ are characterised by great secretory activity, and this is associated with a much-lobed and branched nucleus. The glandular

* Raciborski, M., 'Flora,' vol. 82, pp. 107–132 (1896).

† Meckel, H., 'Archiv f. Anat. Phys. u. Wiss. Med.' (Müller), pp. 1–73 (1846).

‡ Zaddach, G., 'Untersuchungen über die Entwickelung und den Bau der Gliederthiere: I. Die Entwickelung des Phryganideneies,' Berlin, 1854.

§ Korschelt, E., 'Zoologische Jahrbücher: Abtheilung für Anatomie u. Ontogenie der Thiere,' vol. 4 (Part I, 1889), pp. 1–154 (1891).

cells of the parasitic Copepod *Lernanthropus* possess similarly lobed nuclei. The Malpighian tubes of insects are also lined with cells possessing lobed nuclei.

The so-called "nurse-cells" of many insect ovaries form another striking illustration of cells engaged in intense metabolic activity, in which the nuclei have enlarged their surface by intricate lobing (Korschelt).* In the secretory cells of the mammary gland investigated by Nissen† the same end is reached by the actual division of the nucleus into two or three daughter nuclei, without an accompanying cell-division at first taking place.

Among plants we also find records in the literature of similar facts. The tapetal cells of many stamens form a good instance of this kind. In certain pollen grains, at the time when their cytoplasm is undergoing a rapid increase in quantity, the tube-nucleus becomes amœboid in form.

In *Utricularia* haustoria are formed at both ends of the embryo sac, while part of the placenta, near the base of the ovule, becomes modified as a nutritive tissue, which is tapped by the micropylar haustorium. In *U. oligosperma* this placental nutritive tissue is immensely developed, and shows lobed nuclei and some cells with two nuclei.‡ Here, again, we find the association of lobed nuclei or of several free nuclei with a tissue in which active metabolic processes are certainly taking place.

Altogether there is a considerable body of facts all pointing to the importance of the establishment of as large an area of contact as possible between nucleus and cytoplasm during periods of marked metabolic activity. This end is attained either by the lobing of a single nucleus or through the development of a multinucleate condition.

In another way the establishment of a plurality of nuclei by repeated mitotic divisions may aid the interchange of materials between nucleus and cytoplasm. At each division the nuclear membrane disappears, and the karyolymph mingles directly with the cytoplasm. By this means, materials elaborated within the nucleus rapidly pass into the cytoplasm and can be utilised in cell metabolism. In this connection we may recall the case of the Ascidian *Cynthia partita.* Here the nuclear sap in the developing egg escapes into the cytoplasm, where it becomes located in a definite region; in the course of development it is confined to certain cells which ultimately form the ectoderm. The ectoderm thus "owes its origin to the nuclear sap."

* Korschelt, E., 'Zoologische Jahrbücher : Abtheilung für Anatomie u. Ontogenie der Thiere,' vol. 4 (Part I, 1889), pp. 1–154 (1891).

† Nissen, F., 'Archiv f. Mikr. Anat.,' vol. 26, pp. 337–342 (1886).

‡ Merz, M., 'Flora,' vol. 84 (Ergänzungsband), pp. 69–87 (1897).

In *Cerebratulus,* also, the whole quality of the cytoplasm of the developing egg is altered by the escape of nuclear substance into it.*

In several of the plants studied by us, notably in the case of *Tropæolum majus,* nuclear degenerations take place at a very early period whilst mitotic divisions of other nuclei are still actively proceeding. It is possible that such a process of continuous nuclear dissolution, accompanied by repeated nuclear multiplication through the growth and division of the remaining nucleus or nuclei of the cell, may contribute to the cytoplasm material which is of importance in cell metabolism.

It appears probable, therefore, that in some or in all of the ways mentioned above, the multinucleate condition is of direct or indirect value to the plant. The multinucleate state may have arisen, in the first place, merely as a chance incident in the transition from young to adult tissue, owing to the higher susceptibility of the cytoplasm to the altering conditions. But once it made its appearance, it would conceivably afford the organism a distinct advantage in carrying out the chemical processes associated with growth, and might tend to become perpetuated as a definite physiological phase in the history of growing members.

EXPLANATION OF PLATE.

(All drawings were made with the aid of a camera lucida. Leitz's 2·50 mm. oil immersion lens was used with various eye-pieces, except in the case of figs. 27, 28 and 29, in which Zeiss's 2 mm. oil immersion lens and C.0.6 were used. The magnifications given below have been reduced to $\frac{1}{2}$ in reproduction.)

Figs. 1 and 2.—Phragmospheres of *Zea Mays*, L. ($\times$ 800.)

Fig. 3.—Multinucleate cells from leaf-sheath of *Zea Mays* seedling, 1 inch high. ($\times$ 800.)

Fig. 4.—Multinucleate cell from stem of *Zea Mays*, just above first node. ($\times$ 800.)

Fig. 5.—Binucleate rudiment of a vessel from the root of *Zea Mays*, just behind growing point. ($\times$ 800.)

Fig. 6.—One degenerating and one functional nucleus in one cell of *Zea Mays*; stem about 5 mm. above the ninth node. ($\times$ 2000.)

Fig. 7.—Nucleus in prophase of division and another in resting state in a cell of *Anthriscus sylvestris*, Hoffm. ($\times$ 2000.)

Fig. 8.—Cell from stem of *Anthriscus sylvestris* with one phragmosphere, one nucleus in anaphase of division, and one nucleus in resting condition. ($\times$ 800.)

Figs. 9, 10, 11 and 12.—Stages in development of phragmosphere of *Anthriscus sylvestris*. ($\times$ 2000.)

Fig. 13.—Multinucleate cell from stem of *Anthriscus sylvestris*. ($\times$ 800.)

Fig. 14.—One of two nuclei from a cell in the older region of stem of *Anthriscus sylvestris*. ($\times$ 2000.)

Fig. 15.—Four nuclei in a cell of second internode of young plant of *Anthriscus sylvestris*. Three nuclei are still active and one is degenerating. ($\times$ 2000.)

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* MacBride, E. W., "Pres. Add. to Section D (Zoology)," 'Brit. Ass. Adv. Sci. Rep., 86th Meeting,' Newcastle-on-Tyne, pp. 403–417 (1917 for 1916).

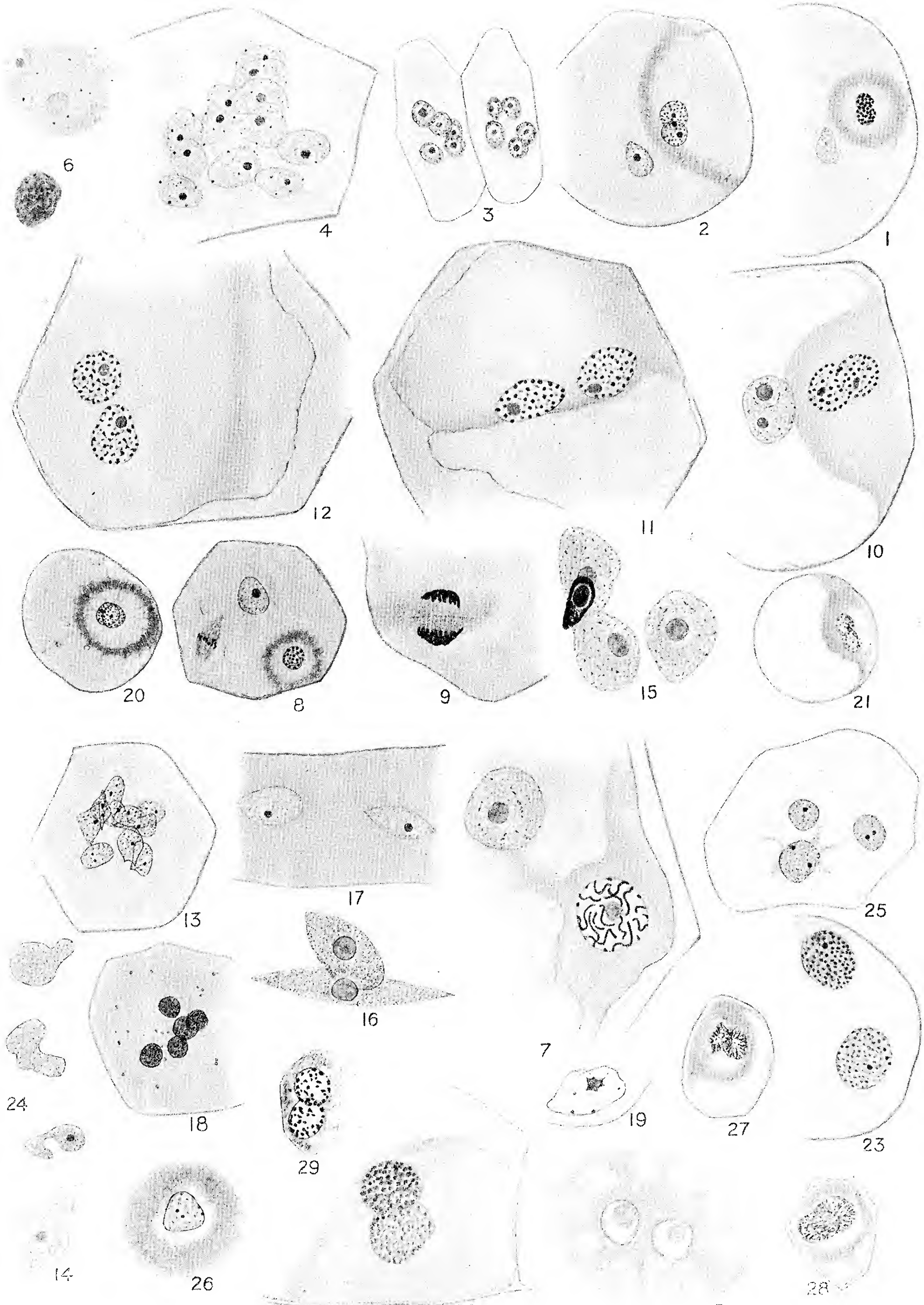
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Fig. 16.—*Rosa* (garden var.). Binucleate cell from young stem. (×2000.)
Fig. 17.—*Rosa* (garden var.). Binucleate cell from stem two years old. (×2000.)
Fig. 18.—*Avena sativa*, L. Parenchyma cell of leaf-sheath. (×800.)
Fig. 19.—*Tropæolum majus*, L. Nucleus from parenchyma of an old stem. (×800.)
Fig. 20.—Phragmosphere from inflorescence axis of *Acanthus mollis*, L., 1 cm. from apex. (×800.)
Fig. 21.—Phragmosphere from stem of *Tropæolum majus*, L. (×800.)
Fig. 22.—Phragmosphere from epidermal cell of leaf base of *Anthurium violaceum*, Schott. (×2000.)
Fig. 23.—Binucleate cell from leaf base of *Larix europæa*, DC (*decidua*). (×2000.)
Fig. 24.—Lobed nuclei from sporangiophore of *Helminthostachys zeylanica*, Hook. (×2000.)
Fig. 25.—Multinucleate cortical cell from sporangiophore of *Equisetum maximum*, Lmk. (×800.)
Fig. 26.—Phragmosphere in a cell from the pith of a stem of *Æsculus Hippocastanum*, L. One of the two daughter nuclei lay exactly beneath the other one, and is not shown in the drawing. (×2000.)
Fig. 27.—*Cryptomeria japonica*, D. Don. Phragmosphere with paired nuclei from a young leaf base. (×1070.)
Fig. 28.—*Lygodium japonicum*, Sw. Cell from mesophyll of climbing petiole, showing two nuclei in a phragmosphere. (×1070.)
Fig. 29.—*Stratiotes aloides*, L. Cell of root cortex from region near apex, showing two nuclei in a phragmosphere. (×1070.)

Concerning Emotive Phenomena.—Part II. *Periodic Variations of Conductance of the Palm of the Human Hand.*

By A. D. Waller, M.D., F.R.S.

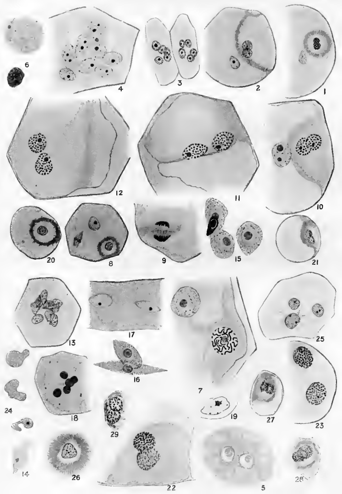
(Received June 7, 1918.)

Introductory.

The present communication is in continuation of that made to the Royal Society on November 8 of last year.

At an early stage of the enquiry, I noticed that the emotive effect varied considerably in magnitude upon the same subject (G. de D.) according as she was rested or fatigued, and that it was usually larger during the day than late in the evening; also that the electrical resistance was higher during the evening than during the day.

I also noticed on my own hand that the electrical resistance was much higher during the night than during the day, *e.g.*, 200,000 ohms at 2 A.M. as compared with 30,000 ohms at 2 P.M., and that the electrical effects on the palm that accompany natural physiological discharges such as coughing, sneezing, and shouting, as well as the natural responses to a pin prick or burn, were remarkably small. (At such time of depression, however, the electrical effect of yawning continued to be exceptionally distinct.)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